REMARKS

Applicants have amended the specification to delete the claim of priority to US Application 09/380137 filed 8/25/1999, which is the National Stage filed under 35 U.S.C. §371 of PCT Application PCT/US99/12252 filed 6/2/1999, which claims priority under 35 U.S.C. §119 to US Provisional Application 60/088863 filed 6/11/1998.

Claims 6-17 are presented for examination. Applicants acknowledge the withdrawal of the rejection of claims 3-17 under 35 U.S.C. § 102(e) as being anticipated by Starling. Applicants thank the Examiner for her review of the instant application. For the reasons stated below, the rejections of the presently pending claims are respectfully traversed.

Priority

The PTO has stated that the subject matter defined in the present application is not supported by the disclosure in any of the applications for which Applicants claim priority because the claimed subject matter does not have utility, enablement, or written description. The PTO therefore sets the priority under 35 U.S.C. §120 to the instant filing date, May 2, 2002.

Applicants have amended the priority information for the instant application herein. Applicants submit that the data in Example 18 (Tumor Versus Normal Differential Tissue Expression Distribution), relied on in part for the utility of the claimed polypeptides, were first disclosed in PCT Application PCT/US00/23328 filed 8/24/2000, on page 93, line 3, through page 96, line 35. For the reasons detailed below, Applicants submit they are entitled to the priority date of August 24, 2000.

Rejection Under 35 U.S.C. §101 – Utility

The PTO maintains its rejection of the pending claims under 35 U.S.C. § 101 as lacking a specific and substantial asserted utility or a well established utility. The PTO states that the specification fails to disclose enough information about the invention to make its usefulness immediately apparent. The PTO also states that Applicants' evidence that differential expression of PRO1138 mRNA in tumor tissue relative to normal tissue is insufficient evidence that the claimed PRO1138 polypeptide will function as a cancer diagnostic.

For the reasons set forth below, Applicants respectfully disagree.

Applicants incorporate by reference their previously submitted arguments, and for the reasons of record assert that the specification contains a disclosure of utility which corresponds in scope to the subject matter sought to be patented and therefore must be taken as sufficient to satisfy the utility requirement of 35 U.S.C. § 101. Applicants also submit that for reasons of record, the PTO has not met its burden of providing evidence showing that one of ordinary skill in the art would reasonably doubt the asserted utility. However even if the PTO has met its initial burden, Applicants' rebuttal evidence previously submitted and additional evidence submitted herewith is sufficient to prove that it is **more likely than not** that a person of skill in the art would be convinced, **to a reasonable probability,** that the asserted utility is true.

As stated previously, Applicants' evidence need not be direct evidence, so long as there is a reasonable correlation between the evidence and the asserted utility. Even if the correlation between Applicants evidence and the asserted utility is not exact, such that there are exceptions to the correlation between the evidence and the asserted utility, this is sufficient to establish a utility. *See Fujikawa v. Wattanasin*, 93 F.3d 1559, 39 U.S.P.Q.2d 1895 (Fed. Cir. 1996) (stating that "a 'rigorous correlation' need not be shown in order to establish practical utility; 'reasonable correlation' suffices," and thus a utility was established even though there were exceptions to the correlation between the disclosed *in vitro* data and asserted *in vivo* utility). Therefore, exceptions between the evidence disclosed and the asserted utility is permissible – **the standard is not absolute certainty.**

Substantial Utility

Summary of Applicants' Arguments and the PTO's Response

Applicants' asserted utility rests on the following argument:

1. Applicants have provided reliable evidence that mRNA for the PRO1138 polypeptide is expressed at least two-fold higher in esophageal and kidney tumors as compared to normal esophageal and kidney tissue, respectively;

2. Applicants assert that it is well-established in the art that differential expression levels of an mRNA for a particular protein, e.g. higher in tumor vs. normal, generally leads to corresponding differential expression levels of the encoded protein, e.g. higher in tumor vs. normal;

3. Given Applicants' evidence that the mRNA for the PRO1138 polypeptide is differentially expressed in esophageal and kidney tumor as compared to normal esophageal and kidney tissue, respectively, it is more likely than not that the PRO1138 polypeptide is similarly differentially expressed in esophageal and kidney tumor as compared to normal esophageal and kidney tissue, respectively, making the claimed polypeptides useful as diagnostic tools, alone or in combination with other diagnostic tools.

Applicants understand the PTO to be making essentially two arguments in response to Applicants' asserted utility:

1. Relying on Hu and LaBaer, the PTO states that "the art clearly teaches that changes in mRNA levels can be either tumor-dependent or tumor-independent," *Office Action* at 3;

2. The PTO argues that "mRNA levels do not correlate with corresponding protein levels," citing a number of references.

Applicants respectfully submit that in light of all of the evidence, the PTO's arguments are not adequate to support the utility rejection of the claimed invention under 35 U.S.C. § 101.

The PTO has Concluded that the data in Example 18 are Sufficient to Establish the Utility of the Claimed Invention

As an initial matter, Applicants point out that in other applications filed by Applicants that rely on data from the exact same disclosure, Example 18, and in which the Applicants have submitted substantially the same references in support of their asserted utility, the PTO has concluded that:

> Based on the totality of evidence of record, **one of skill in the art would find it more likely than not that an increase in message as measured by RTPCR would be predictive of an increase in protein expression levels,** absent evidence to the contrary. Therefore, the data presented in Example 18, which demonstrates differential expression of nucleic acids encoding PRO1180, also supports a conclusion of differential expression of PRO1180 polypeptide. Therefore, one of ordinary skill in the art would be able to use the PRO1180 polypeptide diagnostically for distinguishing normal kidney and rectal tumor tissues compared to kidney tumor and normal rectal tissue, as asserted by Applicant. *Examiner's Reasons for Allowance, Application No. 10/063,529* (emphasis added).

See also *Examiners Reasons for Allowance* in Application No. 10/063,530, No. 10/063,524, No. 10/063,582, and No. 10/063,583, all of which conclude that the data presented in Example 18,

which demonstrate differential expression of the nucleic acids encoding certain PRO polypeptides, also support a conclusion of differential expression of the PRO polypeptides, making the claimed PRO polypeptides and antibodies that bind the PRO polypeptides useful for diagnostic purposes.

Applicants therefore request that the Examiner recognize the utility of the claimed invention, supported by the data presented in Example 18 and the numerous cited references, as was done in the other applications referenced above.

Applicants have established that the Gene Encoding the PRO1138 Polypeptide is Differentially Expressed in Certain Cancers compared to Normal Tissue

Applicants submit that the gene expression data provided in Example 18 of the present application are sufficient to establish that the PRO1138 gene is differentially expressed in esophageal and kidney tumor tissue as compared to their normal tissue counterparts, and is therefore useful as a diagnostic tool for esophageal or kidney cancers. This assertion is based on the results of RT-PCR analysis of pooled normal esophageal or kidney tissue and pooled esophageal or kidney tumor tissue using methods that are well-established in the art.

This utility is substantial, *i.e.* distinguishing tumor cells from normal cells is not an insubstantial or trivial utility without a real world use, and it is specific, *i.e.* it is directed to specific disease and is not a utility that the entire class of nucleic acids shares. Finally, this asserted utility is credible, as one of skill in the art would readily believe that a nucleic acid sequence can be used as a marker to distinguish tumor tissue from normal tissue.

Applicants remind the Examiner that Applicants enjoy a presumption that their assertions are true. The Examiner must approach Applicants' assertion of utility as being sufficient to satisfy the utility requirement. M.P.E.P. §2107.02, "Procedural Considerations Related to Rejections for Lack of Utility," states:

> As a matter of Patent Office practice, a specification which contains a disclosure of utility which corresponds in scope to the subject matter sought to be patented must be taken as sufficient to satisfy the utility requirement of § 101 for the entire claimed subject matter unless there is a reason for one skilled in the art to question the objective truth of the statement of utility or its scope. *M.P.E.P. §2107.02* at III. A., *quoting In re Langer*, 503 F.2d 1380, 1391, 183 USPQ 288, 297 (C.C.P.A. 1974) (emphasis in original).

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> Thus, *Langer* and subsequent cases direct the Office to presume that a statement of utility made by an applicant is true. ... Office personnel should not begin by questioning the truth of the statement of utility. Instead, any inquiry must start by asking if there is any reason to question the truth of the statement of utility. ... Clearly, Office personnel should not begin an evaluation of utility by assuming that an asserted utility is likely to be false, based on the technical field of the invention or for other general reasons. *Id.*

With respect to the use of the PRO1138 nucleic acid to distinguish tumor from normal tissue, the Examiner must accept this assertion as true "unless there is a reason for one skilled in the art to question the objective truth of the statement of utility." Therefore, the question is whether the PTO has established that there is a reason to doubt the objective truth of Applicants' assertion that using standard RT-PCR procedures to examine the expression of the PRO1138 mRNA in pooled samples of esophageal or kidney tumor tissue and pooled samples of corresponding normal tissue, Applicants discovered that PRO1138 mRNA is differentially expressed between normal tissue and tumor tissue such that it can be used as a diagnostic tool.

In response to Applicants' asserted utility, the PTO asserts that "the skilled artisan would not know if the difference in mRNA expression is tumor-dependent or tumor-independent." *Office Action* at 8. This assertion is based on three sentences from a letter to the editor by LaBaer about the Hu reference, and a related statement in the Hu *et al.* reference, of which LaBaer was the primary investigator:

> In the accelerating quest for disease biomarkers, the use of high-throughput technologies, such as DNA microarrays and proteomics experiments, has produced vast datasets identifying thousands of genes whose expression patterns differ in diseased versus normal samples. Although many of these differences may reach statistical significance, they are not always biologically meaningful. For example, reports of mRNA or protein changes of as little as two-fold are not uncommon, and although some changes of this magnitude turn out to be important, most are attributable to disease-independent differences between the samples. *LaBaer* at 976.

> It is not uncommon to see expression changes in microarray experiments as small as 2-fold reported in the literature. Even when these expression changes are statistically significant, it is not always clear if they are biologically meaningful. *Hu* at 411, right column, first full paragraph.

Thus, the PTO is arguing that because "high throughput technologies, such as DNA microarrays" produce differences in mRNA that are attributable to "disease-independent differences between samples," this establishes "a reason for one skilled in the art to question the objective truth" of Applicants' asserted utility which is based on RT-PCR analysis of pooled samples of normal and tumor tissue, not microarrays. Based on Hu and LaBaer, the PTO repeatedly makes the statement, or one very similar, that "the facts to be established are, is the reported change in PRO transcripts tumor-dependent or tumor-independent." *Office Action* at 11, 22, 24, and 25.

Applicants disagree that there is any need to establish the fact that the differential PRO1138 mRNA expression is "tumor-dependent." Applicants' assertion that the RT-PCR data of Example 18 can be used to distinguish tumor from normal tissue, enjoys a presumption of truth, and there are no further "facts" to be established. The issue is whether or not the PTO, relying on Hu and LaBaer, has provided a basis to doubt Applicants' assertions regarding the disclosure in Example 18.

Applicants respectfully submit that one of skill in the art would not accept that the PTO has established a basis to doubt Applicants' assertions. As Applicants' have previously stated, those of skill in the art recognize that RT-PCR is a more accurate and reliable technique than microarrays (see, e.g., Kuo *et al.*, (Proteomics 2005; 5(4):894-906), previously submitted). Therefore, it would be readily apparent to one skilled in the art that opinions regarding data from high-throughput techniques such as microarrays are simply not relevant to Applicants' RT-PCR data, and are not a reason to doubt the truth of Applicants' asserted utility. Thus, even if accurate, a point which Applicants do not concede, Hu's and LaBaer's opinions regarding microarray studies are not relevant to the utility of the instant application which does not rely on microarray data, and cannot form the basis of a reason to doubt Applicants' assertions.

Applicants emphasize that they are <u>not</u> asserting that microarray data are not reliable (that is apparently the PTO's position based on Hu and LaBaer), merely that Applicants are using a method that is recognized by those of skill in the art as more reliable and sensitive.

In response to Applicants' previous arguments based on Kuo, the PTO states:

> Applicants argue Kuo et al that there is a good correlation between mRNA levels and protein levels when the mRNA is measured by quantitative PCR analysis. This is not persuasive, the quantitative levels have not been reported in the

> specification. Further, the studies of Kuo et al differ from the instant situation, because they compare within the same cell, whereas in the instant case, two different cells are compared. There is no evidence of tumor-dependent versus tumor-independent changes. *Office Action* at 16.

The PTO's arguments not only mischaracterize the Applicants' arguments, they completely miss the point of Applicants' reliance on Kuo. Kuo is cited as evidence to support Applicants' assertion that Applicants' PCR data are more accurate and reliable than the microarray technique commented on by Hu and LaBaer. Kuo supports this assertion because it is evidence that one of skill in the art would regard PCR as a more accurate and reliable method of assessing changes in mRNA. Thus, whether or not the microarray technique commented on by Hu and LaBaer yields "disease-independent" results is not relevant to Applicants' data because, as evidenced by Kuo, PCR data such as Applicants' are more accurate and reliable than the microarray data relied on by Hu and LaBaer. Until the PTO provides evidence that transcript changes detected by PCR analysis of pooled normal and tumor samples are often "disease-independent," the PTO's rejection of the data in Example 18 based on Hu and LaBaer is misplaced, and Applicants' asserted utility must be presumed true.

As to the PTO's specific "arguments" in response to Kuo, Applicants note that they are simply statements, not actual argument. The PTO states: "This is not persuasive, the quantitative levels have not been reported in the specification." Applicants agree that the RT-PCR data in Example 18 are not reported in quantitative levels, but fail to see the relevance since the PTO makes no argument based on that fact. Likewise, the PTO states "the studies of Kuo et al differ from the instant situation, because they compare within the same cell, whereas in the instant case, two different cells are compared. There is no evidence of tumor-dependent versus tumor-independent changes." Again, even if true, Applicants fail to see the relevance of this "fact," since the PTO has not offered any explanation of why this "difference" is relevant to Applicants' arguments based on Kuo.

In the same "argument," the PTO states that "Applicants essentially argue that because the PCR analysis is more sensitive at the mRNA level.. then it necessarily follows that protein levels are corresponding. This is not persuasive; the ability to have a more sensitive detection in changes at the mRNA level does not establish that the change is tumor-dependent or that the protein is correspondingly changed." *Office Action* at 16-17. Applicants' have made no such

argument. Applicants argue that PCR is more sensitive than microarray, such that Hu's and LaBaer's opinion regarding microarray data are irrelevant and cannot provide a reason to doubt Applicants' assertions based on PCR data. Thus, the presumption of truth remains. As to the correlation between differential mRNA levels and protein levels, Applicants have cited over 100 references and expert declarations to support this assertion, as is discussed below.

Applicants also note that neither Hu nor LaBaer cite any references to support their assertions that "most [microarray differences] are attributable to disease-independent differences between the samples" and that "it is not always clear if [the microarray differences] are biologically meaningful." In the absence of any supporting references, Applicants cannot independently evaluate these statements to determine what is meant by "disease-independent differences" and "biologically meaningful." Read in light of the entire article and accompanying letter to the editor, Applicants assert that these statements should be interpreted to mean that the observed differences do not play a role in the development or progression of the disease state, or that such a role in the disease state has not yet been published. As Applicants have previously stated, a differentially expressed mRNA can serve as a marker of a disease even if it is "disease-independent" in the sense that it has no role in the cause or progression of a disease, or if any such role is not yet published in the literature. Applicants invite the PTO to provide support for an alternate interpretation of "disease-independent" as used in Hu and LaBaer.

With respect to Applicants' arguments that Hu and LaBaer are silent regarding the reliability of pooled samples, which are of record, the PTO states:

> The asserted diagnostic utility of the PRO polypeptide depends upon its ability to differentiate normal tissue and tumor tissue. In practicing the invention some value for PRO polypeptide expression must be obtained in order to make this distinction. Establishing a cutoff value for this distinction would be difficult unless one knows the degree of variation within the pool, which Applicants have not prodded [*sic*]. There is no evidence of record concerning the normal range of PRO mRNA levels or PRO polypeptide levels in normal tissue or tumor tissue. There is no evidence of record that a normal range of PRO mRNA or PRO polypeptide levels could be defined that would distinguish normal tissue and tumor tissue. Without any knowledge of the variation within the pool one would not know if any particular measurement from a tissue would indicate normal tissue or tumor tissue. Pooled samples would also obscure the variation between samples, making the disclosed results for PRO polynucleotide expression less useful, accurate and informative than if results from individual samples had been provided. In fact the range of values from normal and/or tumor tissue could be so

broad that it would be impossible to distinguish normal tissue from tumor tissue. *Office Action* at 15-16.

The PTO presents no evidence to support these assertions. Thus, the PTO uses conclusory and unsupported arguments as the basis for dismissing the declaration of an expert. As such, the PTO's position is inconsistent with the Utility Examination Guidelines which state, "Office personnel must accept an opinion from a qualified expert that is based upon relevant facts whose accuracy is not being questioned; it is improper to disregard the opinion solely because of a disagreement over the significance or meaning of the facts offered" (*66 Fed. Reg. 1098, Part IIB (2001)*) and also is inconsistent with the requirement of the PTO to support its assertions of fact. *See In re Zurko*, 258 F.3d 1379, 1385, 59 USPQ2d 1693, 1697 (Fed. Cir. 2001). Absent supporting evidence, it is inappropriate for the PTO to dismiss Applicants' arguments and Mr. Grimaldi's opinion regarding pooled samples simply because the PTO wishes to take a contrarian position on the use of pooled samples in discovery of diagnostics.

Regarding the substance of the above-quoted text from the PTO regarding pooled samples, Applicants traverse this position and maintain that their expert has established that "[d]ata from pooled samples is more likely to be accurate than data obtained from a sample from a single individual." *First Grimaldi Declaration* at ¶5. As to the PTO's statement that "[i]n fact, the range of values from normal and/or tumor tissue could be so broad that it would be impossible to distinguish normal tissue from tumor tissue," (*Office Action* at 16 , emphasis added), Applicants note that the Grimaldi declaration make clear that, in fact, "the results of the gene expression studies indicate that the genes of interest can be used to differentiate tumor from normal." *First Grimaldi Declaration* at ¶7. Applicants refrain from further rebutting the PTO's assertions because there presently are no facts on the record to support a position other than that of Mr. Grimaldi's. Applicants respectfully request that the PTO provide evidentiary support for its assertions regarding pooled samples in order to fully develop these issues under examination.

As for the PTO's statement that the first Grimaldi declaration is "in contrast with the specification's teachings," (see *Office Action* at 15), Applicants do not know how to respond since the Office has not explained how the declaration is in contrast with the quoted portion of the specification or what relevance any contrast between the two statements has to Applicants' asserted utility. Similarly, the Office's statement that "Hu and LaBaer are evidence that a skilled

artisan would consider the precise level of PRO gene expression as relevant" (see *Office Action* at 15) is not supported by any reasoning or citation to Hu and LaBaer. Applicants' are unaware of any teaching in Hu and LaBaer regarding the need for a "precise level of PRO gene expression" to use it as a molecular marker to distinguish tumor tissue from normal tissue. In fact, Hu and LaBaer teach nothing at all regarding developing diagnostic markers of cancer.

In conclusion, Applicants submit that the evidence reported in Example 18, supported by the first Grimaldi Declaration, establish that there is at least a two-fold difference in PRO1138 mRNA between esophageal and kidney tumor tissue as compared to their normal tissue counterparts. Applicants' assertion that PRO1138 mRNA can be used to distinguish esophageal and kidney tumor tissue from their normal tissue counterparts must be presumed true by the Examiner unless there is a reason that one of skill in the art would doubt the objective truth of Applicants' statements. Applicants have shown that the references by Hu and LaBaer are inapplicable to Applicants' RT-PCR data, and the PTO has provided no evidentiary basis for dismissing the Grimaldi Declaration. Thus, any challenge to the sufficiency of the data with respect to the utility of the nucleic acid is inappropriate.

Therefore, the only issue which remains is whether the data in Example 18 regarding differential expression of the PRO1138 mRNA are reasonably correlated with differential expression of the PRO1138 polypeptide such that the claimed polypeptides have utility as diagnostic tools as well. As discussed below, even if the PTO has established a reasonable doubt regarding Applicants' assertion that they are reasonably correlated, Applicants' overwhelming rebuttal evidence is more than sufficient to establish that differential mRNA expression leads to corresponding differential expression at the protein level.

The PTO's Evidence is Not Relevant to Determining Whether a Change in mRNA Level for a Particular Gene leads to a Corresponding Change in the Level of the Encoded Protein

Applicants turn next to the second portion of their argument in support of their asserted utility – that it is well-established in the art that a change in the level of mRNA encoding a particular protein generally leads to a corresponding change in the level of the encoded protein; given Applicants' evidence of differential expression of the mRNA for the PRO1138 polypeptide in esophageal and kidney tumors as compared to their normal tissue counterparts, it is likely that

the PRO1138 polypeptide is also differentially expressed; and proteins differentially expressed in certain tumors have utility as diagnostic tools. As stated above, the Examiner should approach these assertions of utility with a presumption that they are true.

In response to Applicants' assertion, the PTO cites, among others, Pennica *et al.*, Gokman-Polar *et al.*, Haynes *et al.*, Gygi *et al.*, Anderson and Seilhamer, and Chen *et al.*; as well as new references: Allman *et al.* (Blood, 1996; 87(12):5357-68), Lian *et al.* (Blood, 2001; 98: 513-524), Fessler *et al.* (J. Biol. Chem. 2002; 277:31291-31302), and Greenbaum *et al.* (Genome Biology, 2003; 4:117.1-117.8); and several references relied on by Applicants: Molecular Biology of the Cell, 3rd ed.; Molecular Biology of the Cell, 4th ed.; Genes VI; Polakis Declaration; and Meric for support of its argument that "one skilled in the cancer diagnostic art would not find it 'more likely than not' that the mRNA levels correspond with the protein levels." *Office Action* at 5.

Applicants have previously discussed at length why the Pennica *et al.*, Konopka *et al.*, Gokman-Polar *et al.*, Haynes *et al.*, Gygi *et al.*, Anderson and Seilhamer, and Chen *et al.*, references are not relevant to the issue of whether differential mRNA expression levels for a particular gene lead to corresponding differential expression of the encoded protein. Briefly stated, references such as Pennica and Konopka which the PTO cites to teach that gene copy number does not equate with mRNA number are not relevant because Applicants do not rely on any relationship between gene copy number and mRNA levels. Likewise, references such as Haynes, Gygi, Anderson, and portions of Chen which discuss the correlation between mRNA level and protein level by examining their static levels across different genes are also not relevant – Applicants rely only on the assertion that differential mRNA expression levels generally lead to corresponding differential expression levels in the encoded protein. Applicants' previous arguments made regarding these issues are of record, and will not be repeated here.

However, because the PTO continues to cite these references, Applicants offer the following illustrations in an attempt to further clarify why references which relate to static global levels of mRNA and protein across different genes are not relevant to Applicants' asserted utility.

Haynes, Gygi, Anderson, and portions of Chen all looked for a correlation between the level of mRNA and corresponding protein by plotting a single measurement of mRNA level vs. protein level for a large group of different genes. The only way that such a plot would result in a

significant correlation is if there exists a global ratio between mRNA levels and protein levels common across all genes, i.e., that for every X copies of an mRNA, there are Y copies of the encoded protein, such that the ratio of X:Y is constant across all genes. If such a global ratio existed, then plotting mRNA levels for different genes against their corresponding protein levels would result in a strong correlation. For example, if the global ratio is 2:1, then 100 transcripts of gene X would result in about 50 copies of protein X, 500 transcripts of gene Y would result in about 250 copies of protein Y, and 1000 transcripts of gene Z would result in about 500 copies of protein Z.

This is what Haynes, Gygi, Anderson, and portions of Chen examined. According to the PTO, they did not find a strong correlation. This is because the ratio between transcript copy number and protein level is apparently not the same for all genes. As a result of these findings, the references concluded that protein levels cannot be accurately calculated from mRNA levels, and that "it is evident that the analysis of mature protein products in cells is essential as there are numerous levels of control of protein synthesis, degradation, processing and modification." *Haynes* at 1863, right column, full paragraph 2. Regardless of whether this conclusion is correct or not, it is not contrary to Applicants' assertion, and is not relevant to the question of whether differential mRNA levels for a particular gene lead to corresponding differential expression of the encoded protein.

In contrast, Applicants' asserted utility does not require knowledge of, or even the existence of, a global ratio between mRNA levels and protein levels. Nor do Applicants' assertions require calculation of protein levels based on measured mRNA levels. Unlike Haynes, Gygi, Anderson, and Chen, Applicants are not relying on a single measure of mRNA for a particular gene and then attempting to calculate protein levels based on a global ratio between mRNA and protein levels. Instead, Applicants are relying on <u>differential</u> mRNA expression, where mRNA levels are measured in two different conditions, i.e. tumor and normal. Applicants assert that a <u>difference</u> in mRNA expression level <u>for a particular gene</u> typically leads to a corresponding <u>difference</u> in the expression level of the encoded protein. *See, e.g., First Grimaldi Declaration* at paragraph 7. The Haynes, Gygi, and Anderson references, as well as portions of the Chen reference, are applicable only to a completely unrelated issue – whether a single

measure of mRNA levels can be used to predict protein levels – and therefore, none of the data or conclusions of these references have any bearing on Applicants' assertions.

To exemplify the difference between these references and Applicants' asserted utilities, Applicants offer the following illustration and analogy with the understanding that like all illustrations and analogies, they are not perfect and therefore do not represent any admissions or binding statements regarding Applicants' disclosure or invention.

Haynes, Gygi, Anderson, and Chen discuss whether there is a correlation between a single measure of mRNA and protein level globally, *i.e.* across different genes at a given time. This is equivalent to conducting a hypothetical Experiment 1, where a particular cell type has 100 copies of mRNA for gene X, 200 copies of mRNA for gene Y, and 400 copies of mRNA for gene Z. If there is a global correlation between static mRNA levels and protein levels across genes, the ratio of the amount of proteins X:Y:Z would be approximately 1:2:4. This is essentially what the cited references examined.

In contrast, Applicants are relying on a correlation between differential mRNA expression for a particular gene leading to a corresponding change in the level of the encoded protein when comparing tissues at two different times or conditions. For example, in hypothetical Experiment 2, if gene X has 100 copies of mRNA per cell in condition A (*e.g.* normal), and 200 copies of mRNA for gene X in condition B (*e.g.* tumor), the amount of protein X in condition B would be greater than the amount of protein X in condition A. Similarly, if gene Y and Z have 300 and 500 copies of mRNA per cell in condition A (*e.g.* normal), respectively, and 900 and 1500 copies of mRNA for gene Y and Z in condition B (*e.g.* tumor), respectively, the amount of protein Y and Z in condition B would be greater than the amount of protein Y and Z in condition A, such that there is a correlation between the difference in the level of mRNA and the difference in the level of protein for a particular gene. This correlation is "global" in the sense that it generally applies to all genes.

The above would be true even if there were no global ratio of mRNA to protein which applies to all genes. For example in Experiment 2, if gene X had an mRNA to protein ratio of 2:1, while gene Y had an mRNA to protein ratio of 1:10, and gene Z had a ratio of 5:1, this would not matter for Applicants' assertion. In all three cases, increasing mRNA would lead to increased protein, and thus there is a "global correlation" – that is to say a there is correlation

between differential mRNA expression and differential protein expression for a particular gene, which generally applies to all genes. This is in spite of the fact that if one were to plot mRNA versus protein for genes X, Y and Z for condition A, as was done in the cited references, there would be no "global correlation" between mRNA and protein level across different genes because for gene X with 100 copies of mRNA the amount of protein X would be 50, for gene Y with 300 copies of mRNA the amount of protein Y would be 3000, and for gene Z with 500 copies of mRNA the amount of protein Z would be 100.

The PTO argues that because there is no correlation between levels of mRNA and protein across genes, *i.e.* a global ratio, as illustrated by Experiment 1 and the plot of mRNA to protein in condition A of Experiment 2, one of skill in the art would not expect an increase or decrease in the amount of mRNA for a particular gene to result in a corresponding change in the amount of the encoded protein, as illustrated in Experiment 2. This is simply wrong – there does not need to be a global ratio between mRNA level and protein level examined across different genes, for there to be a correlation between differential mRNA expression and differential protein expression which applies generally to all genes.

For example, Haynes reports that the amount of protein produced by similar levels of mRNA varied by as much as fifty-fold, and that similar amounts of protein were sustained by amounts of mRNA that varied by as much as forty-fold. *Haynes* at 1863, first full paragraph. Based on these results, Haynes concludes that "protein levels cannot be accurately predicted from the level of the corresponding mRNA transcript." *Id.*

This is analogous to a finding that on one gallon of gas, a hybrid car can travel 50 miles but a large truck can only travel 5 miles, or that to travel 50 miles, a hybrid car requires 1 gallon of gas, but a large truck requires 10 gallons. That is to say, there are many things which affect the fuel efficiency of an automobile. Based on these observations, one could conclude that given the lack of a global ratio of gas to miles that applies across all automobiles, and the resulting lack of correlation between the amount of gas in an automobile and the distance it travels, one cannot predict how far an automobile will travel based on the amount of gas in the tank.

Even if true, Haynes' data and conclusions are irrelevant to Applicants' assertion. Regardless of the fact that there are numerous levels of control of protein expression, Applicants assert that, generally speaking, increasing or decreasing the amount of mRNA for a particular

gene will result in a corresponding increase or decrease in the amount of the encoded protein. This is analogous to increasing or decreasing the amount of gas in an automobile – it will travel farther if you add more gas, and not as far with less. The fact that there are many things which affect fuel efficiency and therefore you cannot predict how far an automobile will travel without knowing if it is a hybrid or a large truck is irrelevant – both a hybrid and a truck travel farther on more gas, and not as far on less.

Applicants emphasize, and the PTO will recognize, that these are simplified illustrations to demonstrate the difference between the two issues being examined. However, these illustrations make clear that even if there is no correlation in the first experiment looking at static levels of mRNA and protein across genes, there can still be a correlation between changes in mRNA and protein for a particular gene as examined in the second experiment.

The PTO's Newly Cited References Do Not Support the PTO's Rejection

In addition to the Haynes, Gygi, Anderson, and Chen, the PTO relies on several new references to support its assertion that "the preponderance of the evidence indicates that the state of the art does not provide for reasonable correlation between mRNA and protein abundances." *Office Action* at 6.

á. Lian et al.

The PTO cites Lian *et al.* for the statement that there is a poor correlation between mRNA expression and protein abundance in mouse cells, and therefore it may be difficult to extrapolate directly from individual mRNA changes to corresponding ones in protein levels. *Office Action* at 6.

In Lian, the authors looked at the mRNA and protein levels of genes in a derived promyelocytic mouse cell-line during differentiation of the cells from a promyelocytic stage of development to mature neutrophils following treatment with retinoic acid. *Lian* at Abstract. The level of mRNA expression was measured using 3'-end differential display (DD) and oligonucleotide chip array hybridization, and protein levels were qualitatively assessed following 2-dimensional gel electrophoresis. *Id.* at Abstract, Table 6.

Lian *et al.* used DD and array hybridization to examine the expression of genes 0, 24, 48 and 72 hours after treatment with retinoic acid. *Id.* at 515, col. 1, ¶ 2. Using this information, the

authors constructed a database of mRNA level changes during differentiation of the cell line. *Id.* at 518, col. 2, ¶ 2. Lian *et al.* also examined protein expression at 0 and 72 hours after retinoic acid treatment. Lian reports that they were able to identify 28 proteins which they considered differentially expressed. *Id.* at 521, Fig. 5. Of those 28, only 18 had corresponding gene expression information in the database, and only 13 had measurable levels of mRNA expression. *Id.* at 521, Table 6. The authors then compared the qualitative protein level from the 2-D electrophoresis gel to the corresponding mRNA level, and reported that only 4 genes of the 18 present in the database had expression levels which were consistent with protein levels. *Id.* at 512, col. 1. The authors note that "[n]one of these was on the list of genes that were differentially expressed significantly (5-fold or greater change by array or 2-fold or greater change by DD)." *Id.* at 512, bridge paragraph (emphasis added). Based on these data, the authors conclude "[f]or protein levels based on estimated intensity of Coomassie dye staining in 2DE, there was poor correlation between changes in mRNA levels and estimated protein levels." *Id.* at 522, col. 2, ¶ 2.

These results are not contrary to Applicants' assertion. Applicants emphasize that they are asserting that a measurable change in mRNA level generally leads to a corresponding change in the level of protein expression, not that changes in protein level can be used to predict changes in mRNA level. Based on the authors' criteria, mRNA levels were significantly changed if they were at least 5-fold different when measured using a microchip array, or 2-fold different when using the more sensitive 3'-end differential display (DD). Of the 28 proteins listed in Table 6, only one has an mRNA level measured by microarray which is differentially expressed according to the authors (spot 7: melanoma X-actin, which mRNA changed from 2539 to 341.3, and protein changed from 1 to 3). None of the other mRNAs listed in Table 6 show a significant change in expression level when using the criteria established by the authors for the less sensitive microarray technique.

There is also one gene in Table 6 whose expression was measured by the more sensitive technique of DD, and its level increased from a qualitative value of 0 to 2, a more than 2-fold increase (spot 2: actin, gamma, cytoplasmic). This increase in mRNA was accompanied by a corresponding increase in protein level, from 3 to 6.

Therefore, although the authors characterize the mRNA and protein levels as having a "poor correlation," this does not reflect a lack of a correlation between a change in mRNA level and a corresponding change in protein level. Only two genes meet the authors' criteria for differentially expressed mRNA level, and of those, one apparently shows a corresponding change in protein level and one does not. *Id.* at 521, Table 6. Thus, there is little basis for the authors' conclusion relied on by the PTO that "it may be difficult to extrapolate directly from individual mRNA changes to corresponding ones in protein levels." *Office Action* at 6 (emphasis added).

b. Fessler et al.

The PTO also cites a publication by Fessler *et al.* *Office Action* at 6. Fessler is not contrary to Applicants' asserted utility, and actually supports Applicants' assertion that a change in the level of mRNA for a particular protein generally leads to a corresponding change in the level of the encoded protein. As noted above, Applicants make no assertions regarding changes in protein levels when mRNA levels are unchanged, nor does evidence of changes in protein levels when mRNA levels are unchanged have any relevance to Applicants' asserted utility.

Fessler *et al.* studied changes in neutrophil (PMN) gene transcription and protein expression following lipoplysaccharide (LPS) exposure. Fessler lists in Table VIII a comparison of the change in the level of mRNA for 13 up-regulated proteins and 5 down-regulated proteins. Of the 13 up-regulated proteins, a change in mRNA levels is reported for only 3 such proteins. For these 3, mRNA levels are increased in 2 and decreased in the third. Of the 5 down-regulated proteins, a change in mRNA is reported for 3 such proteins. In all 3, mRNA levels also are decreased. Thus, in 5 of the 6 cases for which a change in mRNA levels are reported, the change in the level of mRNA corresponds to the change in the level of the protein. This is consistent with Applicants' assertion that a change in the level of mRNA for a particular protein generally leads to a corresponding change in the level of the encoded protein.

Regarding the remainder of the proteins listed in Table VIII, in 6 instances, protein levels changed while mRNA levels were unchanged. This evidence has no relevance to Applicants' assertion that changes in mRNA levels lead to corresponding changes in protein levels, since Applicants are not asserting that changes in mRNA levels are the only cause of changes in protein levels. In the final 6 instances listed in Table VIII, protein levels changed while mRNA was noted as "absent." This evidence also has no relevance to Applicants' assertion that changes

in mRNA levels causes corresponding changes in protein levels. By virtue of being "absent," it is not possible to tell whether mRNA levels were increased, decreased or remained unchanged in PMN upon contact with LPS. Nothing in these results by Fessler suggests that a change in the level of mRNA for a particular protein does not generally lead to a corresponding change in the level of the encoded protein. Accordingly, these results are not contrary to Applicants' assertions.

The PTO points to Fessler's statement regarding Table VIII of "a poor concordance between mRNA transcript and protein expression changes." *Office Action* at 6. As is clear from the above discussion, this statement does not relate to a lack of correlation between a change in mRNA levels leading to a change in protein levels, because in 5 of 6 such instances, changes in mRNA and protein levels correlated well. Instead, this statement relates to observations in which protein levels changed when mRNA was either unchanged or "absent." As such, this statement is an observation that in addition to transcriptional activity, LPS also has post-transcriptional and possibly post-translational activity that affect protein levels, an observation which is not contrary to Applicants' assertions. Accordingly, Fessler's results are consistent with Applicants' assertion that a change in mRNA level of for a particular protein generally leads to a corresponding change in the level of the encoded protein, since 5 of 6 genes demonstrated such a correlation, and do not support the PTO's burden to provide evidence showing that one of ordinary skill in the art would reasonably doubt the asserted utility.

c. Greenbaum et al.

The PTO also cites Greenbaum *et al.* (Genome Biology, 2003; 4:117) for support for its arguments, stating that "Greenbaum et al cautions against assuming that mRNA levels are generally correlative of protein levels. The reference teaches (page 117.3, second column) that primarily because of a limited ability to measure protein abundances, researches have tried to find correlations between mRNA and the limited protein expression data, in the hope that they could determine protein abundance levels from the more copious and technically easier mRNA experiments." *Office Action* at 6 (citations omitted).

Like Haynes, Gygi, Anderson, and portions of Chen, Greenbaum does not provide any support for the PTO's position because the authors examined the correlation between mRNA level and protein level by examining levels across different genes. Applicants have explained

above why such measurements are not relevant to Applicants' assertions. As for the references discussed by Greenbaum in the portion of the paper cited by the Examiner (page 117.3, second column and page 117.04, first column), Greenbaum cites three references which allegedly found a poor or no correlation: Anderson and Seilhamer (Electrophoresis 1997; 18:533-537); Lichtinghagen *et al.* (European Urology 2002; 42:398-406); and Chen *et al.* (Mol. and Cell. Proteomics 2002; 1:304-313). In addition, Greenbaum reports a fourth reference which found a strong correlation: Ornstoft *et al.* (Mol. Cell. Proteomics. 2002; 1(1):37-45). The three references cited by Greenbaum are not contrary to Applicants' assertion, the fourth reference supports Applicants' position, and therefore Greenbaum does not offer the PTO any support for its rejection.

Applicants have already addressed the Anderson reference and explained why it is not relevant. Briefly stated, the authors conducted a study that looked at static levels of mRNA and protein across different genes, *i.e.* a global ratio of mRNA to protein. For the reasons discussed above, a lack of correlation between static levels of mRNA and protein across different genes is not relevant to Applicants' assertions, and therefore Greenbaums' statements based on these experiments are irrelevant and do not support the PTO's rejection.

The second reference cited by Greenbaum is Lichtinghagen *et al.*, stating that the reference shows no significant relationship between mRNA and protein for matrix metalloproteinases (MMPs 2 and 9) and the tissue inhibitor of metalloproteinase 1 (TIMP-1) in human prostate cancer. Lichtinghagen examined the level of MMP-2, MMP-9 and TIMP-1 in cancerous and non-cancerous parts of 17 human prostate samples at both the mRNA and protein level. The level of mRNA was determined using RT-PCR, and the level of protein was determined using quantitative zymography and ELISA. Lichtinghagen reports that comparing non-cancerous to cancerous tissue, mRNA levels were decreased for MMP-2, and unchanged for MMP-9 and TIMP-1. *See Lichtinghagen* at Abstract (attached as Exhibit 1). In contrast, looking at the protein level, MMP-2 levels were unchanged, while MMP-9 expression was higher and TIMP-1 levels were lower. *Id.* Thus, Lichtinghagen reports that there was no correlation between mRNA levels and protein levels. *Id.*

First, it is important to note that of the three genes examined, only one (MMP-2) showed any change in mRNA expression levels between cancerous and non-cancerous tissues. While

statistically significant, the change was small (approximately 33% decrease), far less than a two-fold change. It is therefore not surprising that the authors did not see a measurable change in the amount of MMP-2 protein.

For MMP-9 and TIMP-1, the authors report that there was no change in the level of mRNA, but there was a change in protein level. This apparent lack of correlation between mRNA and protein levels is not contrary to Applicants' assertion that a change in mRNA level generally leads to a change in protein level. Applicants are not attempting to predict the level of mRNA based on changes in protein level, and Applicants have not asserted that the only means for changing the level of protein is to change the amount of the encoding mRNA. Therefore a change in protein without a change in mRNA is not contrary to Applicants' assertions.

Second, the authors in Lichtinghagen note that in another study, researchers found a direct correlation between mRNA levels and protein levels for MMP-2 in prostate cancer. *See Lichtinghagen* at 403, col. 2, *citing* Stearns and Wang (Cancer Res. 1993; 53(4):878-83). In the Stearns and Wang reference cited in Lichtinghagen, the authors report differences in MMP-2 mRNA levels between cancerous, benign and normal stromal tissue from human prostate. The authors state that "[e]nzyme-linked immunosorbent assays demonstrated that the amounts of type IV collangenase protein [MMP-2 protein] correlated directly with the mRNA levels in the tumor tissue." *Stearns and Wang* at Abstract (abstract attached hereto as Exhibit 2). Therefore, contrary to the results reported in Lichtinghagen, at least one other study reports a good correlation between changes in mRNA and protein levels for MMP-2 in prostate cancer.

In conclusion, Lichtinghagen is not contrary to Applicants' assertion that generally, a change in mRNA level leads to a corresponding change in protein level. Lichtinghagen reported a single gene where an apparent change in mRNA did not result in a corresponding change in the level of protein. However, the change in mRNA level was very small, and other researchers have reported a direct correlation between mRNA levels and protein levels for the same gene in human prostate samples. The two other genes examined by Lichtinghagen did not show a change in mRNA level, and therefore say nothing about Applicants' assertion. Therefore, Greenbaum's statements based on Lichtinghagen do not support the PTO's rejection.

The third reference cited by Greenbaum is Chen, *et al.* which was discussed at length previously, as well as above. For the previously stated reasons, Chen is not contrary to

Applicants' assertion, and therefore Greenbaum's reliance on Chen cannot support the PTO's rejection.

In contrast to these three references which offer no or very little support for the PTO's position, Greenbaum also cites a reference by Ornoft *et al.* Applicants have previously discussed Ornoft in detail. Briefly stated, the authors found that "[i]n general there was a highly significant correlation ($p<0.005$) between mRNA and protein alterations. Only one gene showed disagreement between transcript alteration and protein alteration." *Id.* at 42, col. 2. The alterations in mRNA and protein included both increases and decreases. *Id.* at 43, Table II. Clearly, a correlation in 39 of 40 genes examined supports Applicants' assertion that changes in mRNA level generally lead to corresponding changes in protein level.

Thus, when considered as a whole, the references cited by Greenbaum actually support Applicants position since the three which report no correlation are either irrelevant or offer no or little support for the PTO's position, and the one which reports a correlation between changes in gene expression and protein expression reports a correlation for 39 out of 40 genes studied.

Finally, Applicants note that to the extent that the PTO insists on relying on references such as Haynes, Gygi, Anderson and portions of Chen where mRNA/protein relationships are examined across different genes, Greenbaum clearly undercuts the PTO's position that there is no correlation between mRNA and protein levels as applied to Applicants' data. When Greenbaum analyzed differentially expressed mRNAs, the authors found a significant correlation between mRNA and protein levels across different genes. The authors state:

> [W]e looked at correlations between mRNA and protein abundance for those ORFs that had varied or steady levels of mRNA expression over the course of the cell cycle. ... Logically, we would assume that those ORFs that show a large degree of variation in their expression are controlled at the transcriptional level – the variability of the mRNA expression is indicative of the cell controlling mRNA expression at different points of the cell cycle to achieve the resulting and desired protein levels. Thus we would expect, and we found, a high degree of correlation ($r=0.89$) between the reference mRNA and protein levels for these particular ORFs; the cell has already put significant energy into dictating the final level of protein through tightly controlling the mRNA expression, and we assume that there would then be minimal control at the protein level. *Greenbaum* at 117.4, last paragraph, to 117.5, paragraph spanning left and right columns.

This clearly supports Applicants, not the PTO, since Applicants' data and declarations show that the PRO1138 mRNA was differentially expressed by at least two-fold. Applicants

emphasize that studies examining a global relationship between mRNA and protein across genes are not relevant to Applicants' assertions. However, to the extent that the PTO disagrees and continues to rely on Haynes, Gygi, Anderson, and Chen, Greenbaum teaches that one of skill in the art would expect a correlation between mRNA and protein levels across different genes for genes which are differentially expressed. Thus, based on the PTO's own flawed reasoning, Greenbaum clearly supports Applicants' assertions.

In conclusion, nothing in Greenbaum is contrary to Applicants' assertion. The one reference relied on by Greenbaum which is most relevant (Orntoft *et al.*) actually supports Applicants' assertion, and Greenbaum's analysis of differentially expressed genes significantly undercuts the PTO's position based on Haynes, Gygi, Anderson, and portions of Chen, since Greenbaum found a high degree of correlation between mRNA levels and protein levels for differentially expressed genes.

d. Allman et al.

The PTO cites Allman *et al.* (Blood, 87(12):5257-68 (1996)) as supporting the assertion that "[d]ifferential analysis of mRNA expression is not always correlated with protein levels." *Office Action* at 7. The PTO states that Allman disclosed that germinal center B cells express dramatically more BCL-6 protein than resting B cells, despite similar BCL-6 mRNA levels in the two cell populations. *Office Action* at 7 and 9. Applicants submit that Allman is not contrary to Applicants' asserted utility because Allman does not teach that a change in mRNA level does not lead to a corresponding change in level of the encoded polypeptide – at best Allman teaches that mechanisms other than increasing mRNA levels can lead to increased protein levels. This is not contrary to Applicants' assertion.

Applicants disclose in Example 18 differential expression of PRO1138 mRNA in esophageal and kidney tumor tissue compared to their respective normal tissue counterparts. Applicants submit that one skilled in the art would expect that a change in mRNA level would generally lead to a corresponding change in the level of the encoded polypeptide. Applicants make no assertions regarding expected changes in protein levels when mRNA levels are unchanged, and evidence of changes in protein levels when mRNA levels are unchanged has no relevance to Applicants' assertion.

Allman does not stand for a position contrary to Applicants' asserted utility. If anything, Allman supports Applicants' utility. Allman states that "BCL-6 protein was readily detectable in germinal center cells (Fig. 6A) and in B-cell lines that express BCL-6 mRNA (Fig. 6A, BJAB and RL), but not in B-cell lines that express little or no BCL-6 mRNA (Fig 6A, VDSO)." *Allman* at 5263, right column. Thus, Allman teaches that for cells expressing higher levels of BCL-6 mRNA, BCL-6 polypeptide levels also were higher, relative to BCL-6 polypeptide levels in cells that expressed lower levels of BCL-6. This is consistent with Applicants' assertions.

Furthermore, Applicants maintain that Allman's discussion of the observed protein levels supports Applicants' assertion that it is well-established in the art that in general, the level of protein is positively correlated to the level of mRNA. In the discussion of their finding that mRNA and protein levels were not correlated, Allman refers to the discovery as a "striking dichotomy." *Allman et al.* at 5265, right column. They also state that "an *unanticipated* finding was that the higher BCL-6 protein levels...could not be fully accounted for by increased mRNA expression." *Allman et al.* at 5267, left column (emphasis added). Both of these statements indicate that normally, protein expression is correlated to mRNA levels, and their findings to the contrary were unexpected for that reason.

In conclusion, Allman does not teach that changes in mRNA level do not lead to corresponding changes in the level of the encoded polypeptide. Accordingly, Allman is not contrary to Applicants' asserted utility and does not support the PTO's position. In fact, Allman provides teachings consistent with Applicants' asserted utility.

The PTO apparently does not understand Applicants' argument that while mRNA changes generally lead to protein changes, not all protein changes are a result of changes in mRNA. The PTO argues:

> If one is to argue, as Applicant has argued, that because PRO transcripts are differentially expressed in tumors it is more likely than not that the PRO polypeptide is similarly differentially expressed in tumors, and therefore PRO polypeptide and if one is to argue, as Applicants have argued, that because PRO transcripts are antibodies can be used for tumor diagnosis [*sic*], then one must also accept the argument that because resting B cells and germinal center B cells express similar BCL-6 mRNA levels it is more likely than not that the BCL-6 protein is not differentially expressed in these two cell populations, and therefore BCL-6 protein and antibodies thereto cannot be used as a marker for germinal center B cells. One must also accept the argument that because germinal center B-cells express dramatically more BCL-6 protein than resting B cells it is more

> likely than not that BCL-6 mRNA is differentially expressed in these two cell populations, and therefore BCL-6 mRNA can be used as a marker for germinal B-cells. Allman indicates that this is not so. *Office Action* at 21.

Applicants are not arguing that a change in polypeptide levels generally causes changes in mRNA levels or that polypeptide levels serve as indicators of mRNA levels. This argument conflates cause and effect. Nor do Applicants argue that a change in mRNA level is the sole cause of changes in the level of the encoded polypeptide. Applicants merely submit that one skilled in the art would expect that a change in mRNA levels for a particular gene would generally lead to a corresponding change in levels of the encoded polypeptide. Allman is consistent with Applicants' contentions because Allman teaches that for cells expressing higher levels of BCL-6 mRNA, BCL-6 polypeptide levels also were higher, relative to BCL-6 polypeptide levels in cells that expressed lower levels of BCL-6 mRNA. Accordingly, Allman does not support a rejection of the claims for lacking utility.

Finally, Applicants address the PTO's repeated reliance on the assertion that:

> In view of the fact that there are numerous levels of control of protein synthesis, degradation, processing, and modification, that are only apparent by direct analysis, the skilled artisan would not know if the disclosed difference in mRNA expression is associated with the corresponding change in the level of protein. *Office Action* at 7; *see also Office Action* at 20-21, 22, and 25-26.

This assertion is based on a statement in Haynes. However, as discussed above, the authors of Haynes based their conclusions on a lack of a global relationship between mRNA levels and protein levels across different genes. This is not relevant to the question of whether one of skill in the art would expect changes in mRNA level to lead to changes in protein level. The authors of Haynes did not address this question, and their statements regarding the ability to calculate protein levels based on mRNA levels refer only to their experiments looking for a global correlation.

In addition to Haynes, the PTO relies on Allman, Molecular Biology of the Cell, 3rd ed., Molecular Biology of the Cell, 4th ed., Genes VI, the Polakis Declaration, and Meric (*see Office Action* at 9, 20-21, 22, and 25-26). Applicants note that the PTO is not considering the entire teachings of these references when it chooses to ignore portions of the text which support Applicants. For example, the PTO cites Genes VI as teaching that "the production of RNA cannot inevitably be equated with production of protein," (*see Office Action* at 9), while the full

statement reads "having acknowledged that control of gene expression can occur at multiple stages, and that production of RNA cannot inevitably be equated with production of protein, it is clear that the overwhelming majority of regulatory events occur at the initiation of transcription." *Genes VI* at 847-848 (emphasis added). How each of these references are either not relevant, or support Applicants' position when read in their entirety, is of record, and will not be repeated here.

However, even if one acknowledges that there are numerous levels of control of protein synthesis, degradation, processing and modification, this is still not contrary to Applicants' assertion that when mRNA levels for a particular gene are changed, there is generally a corresponding change in protein levels. Just because a cell has numerous means of modulating protein levels, this does not prohibit the possibility that a change in mRNA level generally results in change in protein level – these are not mutually exclusive propositions. That is to say that it is possible that if the cell has put significant energy into dictating the final level of protein through tightly controlling mRNA expression, then there could be minimal control at the protein level. *See, e.g., Greenbaum* at 117.4, last paragraph, to 117.5, paragraph spanning left and right columns. Therefore, one must look at actual experiments where a change in mRNA level was assessed to determine if the change generally results in a corresponding change in protein levels. None of the references cited by the PTO teach to the contrary, and Applicants' evidence discussed below teaches that this is in fact the case.

In conclusion, Applicants have shown that the references by Haynes, Gygi, Anderson, Greenbaum and portions of Chen that examine mRNA/protein relationships across different genes are simply not relevant to the issue of whether a change in mRNA levels leads to a corresponding change in the level of the encoded protein. Likewise, the PTO's new references, Lian, Fessler, and Allman, that examine differential mRNA expression are not contrary to Applicants' position. In addition, Applicants have shown that Fessler and Allman are consistent with and actually support Applicants' position, not the PTO's. Taken together, the PTO's arguments are not sufficient to satisfy the burden to "provide[] evidence showing that one of ordinary skill in the art would reasonably doubt the asserted utility." *In re Brana*, 51 F.3d 1560, 1566, 34 U.S.P.Q.2d 1436 (Fed. Cir. 1995).

Applicants' Evidence Establishes that a Change in mRNA Level for a Particular Gene leads to Corresponding Change in the Level of the Encoded Protein

In support of the assertion that changes in mRNA for a particular gene are positively correlated to changes in the corresponding protein level, Applicants previously submitted a copy of a second Declaration by J. Christopher Grimaldi, a copy of the declaration of Paul Polakis, Ph.D., excerpts from the Molecular Biology of the Cell, a leading textbook in the field (Bruce Alberts, *et al.*, Molecular Biology of the Cell (3rd ed. 1994) and (4th ed. 2002), excerpts from the textbook, Genes VI, (Benjamin Lewin, Genes VI (1997)), a reference by Zhigang *et al.*, World Journal of Surgical Oncology 2:13, 2004, and a reference by Meric *et al.*, Molecular Cancer Therapeutics, vol. 1, 971-979 (2002). In addition, in the most recent response, Applicants submitted over 100 additional references in support of their assertion that changes in mRNA for a particular gene are positively correlated to changes in the corresponding protein level. The details of the teachings of these declarations and references, and how they support Applicants' asserted utility, are of record and will not be repeated here.

Applicants submit herewith a copy of a declaration by Randy Scott, Ph.D. (attached as Exhibit 3). Dr. Scott is an independent expert in the field of molecular diagnostics, with over 15 years experience. He is the author of over 40 scientific publications in the fields of protein biology, gene discovery, and cancer, and is an inventor on several issued patents. His curriculum vitae is attached to the declaration. In paragraph 10 of his declaration, Dr. Scott states:

> One reason for the success and wide-spread use of the DNA microarray technique, which has led to the emergence of a new industry, is that generally there is a good correlation between mRNA levels determined by microarray analysis and expression levels of the translated protein. Although there are some exceptions on an individual gene basis, it has been a consensus in the scientific community that elevated mRNA levels are good predictors of increased abundance of the corresponding translated proteins in a particular tissue. Therefore, diagnostic markers and drug candidates can be readily and efficiently screened and identified using this technique, without the need to directly measure individual protein expression levels. *Scott Declaration* at ¶10 (emphasis added).

Applicants submit the opinion of yet another expert in the field that changes in mRNA level for a particular protein in a given tissue generally lead to a corresponding change in the level of the encoded protein. Importantly, Dr. Scott also states that, contrary to the contentions of the PTO, diagnostic markers can be identified "without the need to directly measure individual protein

expression levels." This opinion is supported by Dr. Scott's extensive experience in the field, as well as the fact that an entire industry has developed around technology to assess differential mRNA expression. As stated previously, there would be little reason to study changes in mRNA expression levels if those changes did not result in corresponding changes in the encoded protein levels.

Applicants also submit herewith a copy of a second Declaration by Dr. Polakis (attached as Exhibit 4) that presents evidentiary data in Exhibit B. Exhibit B of the Declaration identifies 28 gene transcripts out of 31 gene transcripts (i.e., greater than 90%) that showed good correlation between tumor mRNA and tumor protein levels. As Dr. Polakis' second Declaration says "[a]s such, in the cases where we have been able to quantitatively measure both (i) mRNA and (ii) protein levels in both (i) tumor tissue and (ii) normal tissue, we have observed that in the vast majority of cases, there is a very strong correlation between increases in mRNA expression and increases in the level of protein encoded by that mRNA." Accordingly, Dr. Polakis has provided the facts to enable the PTO to draw independent conclusions.

The case law has clearly established that in considering affidavit evidence, the PTO must consider all of the evidence of record anew. *See in re Rinehart,* 531 F.2d 1084, 189 USPQ 143 (C.C.P.A. 1976); *In re Piasecki,* 745 F.2d. 1015, 226 USPQ 881 (Fed. Cir. 1985). "After evidence or argument is submitted by the applicant in response, patentability is determined on the totality of the record, by a preponderance of the evidence with due consideration to persuasiveness of argument." *In re Alton,* 37 U.S.P.Q.2d 1578, 1584 (Fed. Cir. 1996), *quoting In re Oetiker,* 977 F.2d 1443, 1445, 24 U.S.P.Q.2d 1443, 1444 (Fed. Cir. 1992). Furthermore, the Federal Court of Appeals held in *In re Alton*, "We are aware of no reason why opinion evidence relating to a fact issue should not be considered by an examiner." *Id.* at 1583. Applicants also respectfully draw the PTO's attention to the Utility Examination Guidelines which state, "Office personnel must accept an opinion from a qualified expert that is based upon relevant facts whose accuracy is not being questioned; it is improper to disregard the opinion solely because of a disagreement over the significance or meaning of the facts offered." *66 Fed. Reg. 1098, Part IIB (2001).*

In summary, Applicants have submitted herewith additional expert Declarations in addition to the declarations and over 115 references already of record, which support Applicants'

asserted utility, either directly or indirectly. This evidence supports the assertion that in general, a change in mRNA expression level for a particular gene leads to a corresponding change in the level of expression of the encoded protein. As Applicants have previously acknowledged, the correlation between changes in mRNA level and protein level is not exact, and there are exceptions. However, Applicants remind the PTO that the asserted utility does not have to be established to a statistical certainty, or beyond a reasonable doubt. *See M.P.E.P.* at § 2107.02, part VII (2004). Therefore, the fact that there are exceptions to the correlation between changes in mRNA and changes in protein does not provide a proper basis for rejecting Applicants' asserted utility. Applicants submit that considering the evidence as a whole, with the overwhelming majority of the evidence supporting Applicants' asserted utility, a person of skill in the art would conclude that Applicants' asserted utility is "more likely than not true." *Id.*

In conclusion, Applicants submit that they have offered sufficient evidence to establish that it is more likely than not that one of skill in the art would believe that because the PRO1138 mRNA is differentially expressed in esophageal and kidney tumor tissue as compared to their respective normal tissue counterparts, the PRO1138 polypeptide will likewise be differentially expressed. This differential expression of the PRO1138 polypeptide makes the claimed polypeptides useful as diagnostic tools for cancer, particularly esophageal or kidney cancer.

The PTO Has Failed to Address Applicants' Evidence of Record

In Applicants' most recent response, Applicants submitted over 100 additional references in support of their assertion that changes in mRNA for a particular gene are positively correlated to changes in the corresponding protein level. The PTO asserts that "the preponderance of the evidence indicates that the state of the art does not provide for a reasonable correlation between mRNA and protein abundances. ... In view of the totality of the evidence of record, one skilled in the art would not assume that gene expression (mRNA) necessarily parallels or is predictive of protein expression." *Office Action* at 6.

However, the PTO has not addressed the over 100 additional references which Applicants have made of record, or even acknowledged that they were submitted. The only possible exception is the statement that "Applicants argue that they assert a change in mRNA levels is reflected in a change of protein levels and rely on a plethora of references to show, where

actually studied that there is a corresponding change. This is not persuasive for reasons of record." *Office Action* at 18.

These "reasons of record" are based on the evidence presented by the PTO which Applicants have addressed above. As discussed, most of the PTO's references are irrelevant, offer no or at best minimal support for the PTO, or actually support Applicants' assertions. The PTO impermissibly uses the teachings of these references to dismiss Applicants' "plethora" of evidence in support of the assertion that changes in mRNA levels are typically accompanied by changes in levels of the encoded polypeptide without ever considering Applicants' evidence.

Applicants submit that this does not represent full consideration of the totality of the record in fairly evaluating the utility of the claimed polypeptides. As provided in the M.P.E.P.:

> It is essential for Office personnel to recognize, fully consider and respond to each substantive element of any response to a rejection based on lack of utility. Only where the totality of the record continues to show that the asserted utility is not specific, substantial, and credible should a rejection based on lack of utility be maintained. *M.P.E.P.* § 2107

Applicants submit that the issue of whether changes in the level of mRNA for a particular gene lead to corresponding changes in the level of the encoded protein requires a determination based on the totality of the record. The totality of the record as it stands is clear: one skilled in the art would reasonably believe that changes in mRNA levels typically lead to corresponding changes in the levels of the encoded protein. There is no basis in PTO policy or the standards set by the courts for the PTO to ignore evidence submitted by Applicants or for the PTO to make an adverse conclusion on the utility of claims based on less than the totality of the evidence. Accordingly, Applicants respectfully request that the PTO consider references of Exhibits 3-13 in particular, as well as Exhibits 14-22, submitted in Applicants' previous response. Applicants submit that, contrary to the PTO's holding, the totality of the record shows that changes in the level of mRNA for a particular gene generally lead to corresponding changes in the level of the encoded protein, and the PTO's holding otherwise does not represent a fair and full consideration of the totality of the record, as required.

The PTO's Position Regarding Specific Evidence of Differential PRO Polypeptide Expression is Inconsistent with the Utility Guidelines and the Courts

In response to Applicants' evidence and arguments, the PTO takes the position that Applicants must present specific evidence directly demonstrating the utility of the claimed polypeptides – specifically, direct evidence of differential expression of PRO1138 polypeptide in tumor and normal tissue. The PTO states:

> Neither the specification nor any of Applicants' arguments, exhibits, declarations or other evidence providing any specific data disclosing if or how the PRO polypeptide expression changes in tumor tissue. Instead, Applicants rely on a general correlation between mRNA expression and expression of the encoded protein rather than the specific correlation between PRO transcripts and PRO polypeptide expression to argue that it is more likely than not that a change in PRO transcripts is correlated with an assumed change in PRO polypeptide expression. Without any evidence of the expression of PRO polypeptide in tumor tissue or normal tissue this argument is of no avail to Applicants. *Office Action* at 9-10 (emphasis added).
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> The specification does not establish if the disclosed change in PRO mRNA expression is one of those cases where there is a correlation between mRNA expression and polypeptide expression. ... In the absence of any testing of the expression of the PRO polypeptide, the specification does not provide some immediate benefit to the public for the PRO polypeptide. ... Instead, Applicants merely propose a utility that is not implausible, relying on a general correlation between mRNA expression and expression of the encoded protein rather than the specific correlation between PRO mRNA expression and PRO polypeptide expression without any evidence of the expression level of the polypeptide in tumor tissue or normal tissue. *Office Action* at 11-12 (emphasis added).
>
> ***
>
> Applicants have not examined whether the reported change in PRO transcripts is correlated with a corresponding change in PRO polypeptide expression. ... Applicants assume that transcript levels are indicative [of] PRO polypeptide levels. The specification fails to provide any testing of PRO polypeptide levels. *Office Action* at 20 (emphasis added).
>
> ***
>
> The fact that there may be a commonly understood general rule or dogma that increased mRNA levels are predictive of corresponding increased levels of the

> encoded protein does not establish the correlation between the change, if any, in PRO transcripts and PRO polypeptide expression in tumors because there are examples of genes for which such a correlation does not exist, as evidenced by the Polakis declaration. *Office Action* at 23 and 25.

Thus, the PTO implies the following argument: (1) the evidence of record demonstrates that there are exceptions to the general rule that increased mRNA levels correspond to increased levels of the encoded polypeptide; (2) because such exceptions exist, it is mandatory that specific data of differential PRO1138 polypeptide expression in esophageal or kidney tumor tissue as compared to their respective normal tissue counterparts be disclosed; and (3) since such direct evidence is not disclosed, the PRO1138 polypeptide has no substantial utility.

Adopting the PTO's standard for utility would result in a per se rule that a difference in mRNA expression cannot establish a utility for the encoded polypeptide and antibodies thereto. Thus, the PTO chooses to heighten the utility requirement to require specific, direct evidence of utility when there are exceptions to a generally accepted rule that is relied upon for utility. This heightened utility requirement is inconsistent with the Utility Guidelines and the courts. There is no requirement that utility be dispositively proven:

> Furthermore, the applicant does not have to provide evidence sufficient to establish that an asserted utility is true "beyond a reasonable doubt." *In re Irons,* 340 F.2d 974, 978, 144 USPQ 351, 354 (CCPA 1965) ... Instead, evidence will be sufficient if, considered as a whole, it leads a person of ordinary skill in the art to conclude that the asserted utility is more likely than not true. *M.P.E.P.* 2107.02 VII (emphasis in original).

Nor is there a requirement that only direct evidence of utility is sufficient to establish utility. Instead, it is established law that indirect evidence that is reasonably indicative of utility is sufficient to fulfill the requirements of 35 U.S.C. §101. *Nelson v. Bowler*, 626 F.2d 853, 856. Furthermore, there is no requirement that indirect evidence necessarily and always prove actual utility. Instead, there only need be a reasonable correlation between the indirect evidence and the asserted utility. *Id.* at 857, *Cross v. Iizuka*, 753 F.2d 1040, 1050-1051. The indirect evidence need not absolutely prove the asserted utility. All that is required is that the tests be reasonably indicative of the asserted utility. In other words, there need only be a sufficient correlation between the indirect evidence and the utility so as to convince those skilled in the art, to a

reasonable probability, that the novel compound will possess the asserted utility. *Fujikawa v. Wattanasin*, 93 F.3d 1559, 1564.

The PTO appears to consider the above guidance from the courts inapplicable to the present situation because in those cases the claimed compound had been tested, and, in the present test, the claimed polypeptides have not been tested. However, the PTO's position fails to recognize the issue in question for the above cases. The issue in question was whether or not Appellants' evidence (*in vitro* or animal testing of compound), which was different in nature from the asserted utility (therapeutic use of compound), was sufficient to fulfill the requirements of 35 U.S.C. §101 when there was a reasonable link between Appellants' evidence and the asserted utility. In the present case, Applicants submit that their evidence (differential mRNA expression) is reasonably linked to the asserted utility (diagnostic use of the encoded polypeptide). Insofar as it is uncontested that differential mRNA expression is reasonably linked to differential polypeptide expression, Applicants submit that such linkage is sufficient to fulfill the requirements of 35 U.S.C. §101 as provided by the guidance of the Utility Guidelines and the courts.

The PTO dismisses the above direction from the Utility Guidelines and the Courts stating:

> Applicants' utility standard would mandate only a showing that it is not implausible, that the invention will work for its intended purpose. If mere plausibility were the test for how to use a claimed invention, Applicants could obtain patent rights to "inventions" based on a disclosure consisting of little more than guesses as to the likelihood of their success. *Office Action* at 10-11 (emphasis added).

Applicants emphasize that it is not "Applicants' utility standard" which is described above, but rather the utility standard established by the PTO's guidelines based on the law as stated by the Courts. The M.P.E.P., citing the relevant case law from the Courts, states that the standard for establishing utility is "more likely than not true":

> [T]he applicant does not have to provide evidence sufficient to establish that an asserted utility is true "beyond a reasonable doubt." *In re Irons,* 340 F.2d 974, 978, 144 USPQ 351, 354 (CCPA 1965). Nor must an applicant provide evidence such that it establishes an asserted utility as a matter of statistical certainty. *Nelson v. Bowler*, 626 F.2d 853, 856-57, 206 USPQ 881, 883-84 (CCPA 1980)... Instead, evidence will be sufficient if, considered as a whole, it leads a person of

> ordinary skill in the art to conclude that the asserted utility is more likely than not true. *M.P.E.P.* 2107.02 VII (emphasis in original).

Because the standard for satisfying the utility requirement is low – more likely than not true – the M.P.E.P. cautions that:

> Rejections under 35 U.S.C. 101 have been **rarely** sustained by federal courts. Generally speaking, in these **rare** cases, the 35 U.S.C. 101 rejection was sustained [] because the applicant ... asserted a utility that could only be true if it violated a scientific principle, such as the second law of thermodynamics, or a law of nature, or was wholly inconsistent with contemporary knowledge in the art. *M.P.E.P.* § 2107.02 III B., citing *In re Gazave*, 379 F.2d 973, 978, 154 U.S.P.Q. 92, 96 (C.C.P.A. 1967) (underline emphasis in original, bold emphasis added).

In conclusion, the PTO's heightened requirement for establishing utility of the presently claimed polypeptides is contrary to the Utility Guidelines and the courts: it is sufficient to present evidence of differential mRNA expression since it is understood in the art that differential mRNA expression is reasonably linked to differential polypeptide expression. As discussed above, even if the PTO has presented evidence that changes in mRNA expression are not always correlated with changes in protein expression, Applicants' overwhelming rebuttal evidence, which has yet to be considered by the PTO, is more than sufficient to establish that changes in mRNA level typically lead to corresponding changes in protein level.

As such, Applicants have established that it is more likely than not that one of skill in the art would believe that because the PRO1138 mRNA is differentially expressed in esophageal and kidney tumor tissue as compared to their respective normal tissue counterparts, the PRO1138 polypeptide will likewise be differentially expressed. Accordingly, when the totality of the evidence is applied to the proper standard for utility, it is clear that this differential expression of the PRO1138 polypeptide establishes the claimed polypeptides are useful as diagnostic tools for cancer, particularly esophageal or kidney cancer.

Specific Utility

The Asserted Substantial Utilities are Specific to the Claimed Polypeptides

Applicants next address the PTO's assertion that the asserted utilities are not specific to the claimed polypeptides related to PRO1138. Applicants respectfully disagree.

Specific utility is defined as utility which is "specific to the subject matter claimed," in contrast to "a general utility that would be applicable to the broad class of the invention." M.P.E.P. § 2107.01 I. Applicants submit that the evidence of differential expression of the PRO1138 gene and polypeptide in certain types of tumor cells, along with the declarations and references discussed above, provide a specific utility for the claimed polypeptides.

As discussed above, there are significant data which show that the gene for the PRO1138 polypeptide is more highly expressed in esophageal and kidney tumor tissue compared to normal esophageal and kidney tissue, respectively. These data are strong evidence that the PRO1138 gene and polypeptide are associated with esophageal and kidney tumors. Thus, contrary to the assertions of the PTO, Applicants submit that they have provided evidence associating the PRO1138 gene and polypeptide with a specific disease. The asserted utility for the claimed polypeptides as diagnostic tools for cancer, particularly esophageal and kidney tumors, is a specific utility – it is not a general utility that would apply to the broad class of polypeptides.

Conclusion

Applicants remind the PTO that the evidence supporting utility does not need to be direct "specific" evidence, nor does it need to provide an exact correlation between the submitted evidence and the asserted utility. Instead, evidence which is "reasonably" correlated with the asserted utility is sufficient. *See Fujikawa v. Wattanasin*, 93 F.3d 1559, 1565, 39 U.S.P.Q. 2d 1895 (Fed. Cir. 1996) ("a 'rigorous correlation' need not be shown in order to establish practical utility; 'reasonable correlation' suffices"); *Cross v. Iizuka*, 753 F.2d 1040, 1050, 224 U.S.P.Q. 739 (Fed. Cir. 1985) (same); *Nelson v. Bowler*, 626 F.2d 853, 857, 206 U.S.P.Q. 881 (C.C.P.A. 1980) (same). In addition, utility need only be shown to be "more likely than not true," not to a statistical certainty. *M.P.E.P.* at § 2107.02, part VII (2004). Considering the evidence as a whole in light of the relevant standards for establishing utility, Applicants have established at least one specific, substantial, and credible utility. In view of the above, Applicants respectfully request that the PTO reconsider and withdraw the utility rejection under 35 U.S.C. §101.

Rejections under 35 U.S.C. § 112, first paragraph – Enablement

The PTO also maintains its rejection of pending Claims 6-17 under 35 U.S.C. § 112, first paragraph. Specifically, the PTO asserts that because the claimed invention is not supported by either a specific or substantial asserted utility or a well established utility, one skilled in the art would not know how to use the claimed invention. *Office Action* at 28.

Applicants submit that in the discussion of the 35 U.S.C. § 101 rejection above, Applicants have established a substantial, specific, and credible utility for the claimed polypeptides. To the extent that the enablement rejection is based on a lack of utility, Applicants respectfully request that the PTO reconsider and withdraw the enablement rejection under 35 U.S.C. §112.

Rejection under 35 U.S.C. §112, first paragraph – Written Description

The PTO also rejects 14-17 under 35 U.S.C. §112, first paragraph, as failing to comply with the written description requirement. Specifically, the PTO alleges that the specification does not provide sufficient distinguishing identifying characteristics of the genus of claimed polypeptides.

The PTO has Failed to Meet Its Initial Burden of Rebutting the Presumption that the Pending Claims are Adequately Described

Claims 14-17 recite the limitation "wherein said isolated polypeptide or a fragment thereof can be used to generate an antibody which can be used to specifically detect the polypeptide of SEQ ID NO:46 in esophageal or kidney tissue samples." In response to Applicants' previous arguments, the PTO argues:

> The specification intends immunologically active peptides to also retain biological activity of a native or naturally-occurring PRO, as indicated below: "Active" or "activity" for the purposes herein refers to form(s) of a PRO polypeptide which retain a biological and/or an immunological activity of native or naturally-occurring PRO, wherein "biological" activity refers to a biological function (either inhibitory or stimulatory) caused by a native or naturally-occurring PRO other than the ability to induce the production of an antibody against an antigenic epitope possessed by a native or naturally-occurring PRO and an "immunological" activity refers to the ability to induce the production of an antibody against an antigenic epitope possessed by a native or naturally-occurring PRO. Page 39, paragraph 0231. Therefore, the claims encompass any and all antigenically cross-

> reactive polypeptides possessing the recited percent identity to the amino acid sequence of SEQ ID NO:46, and possessing any and/or all underlying biological activities. However, the specification does not describe any biological activity of the native or naturally-occurring PRO polypeptide SEQ ID NO:46. *Office Action* at 29-30.

As noted previously, "[a] description as filed is presumed to be adequate, unless or until sufficient evidence or reasoning to the contrary has been presented by the examiner to rebut the presumption." *M.P.E.P.* § 2163.04 (emphasis added). Therefore "[t]he examiner has the initial burden of presenting by a preponderance of evidence why a person skilled in the art would not recognize in an applicant's disclosure a description of the invention defined by the claims." *Id.* The above arguments fail to meet this burden because they are fundamentally flawed for at least two reasons.

First, the PTO is relying on a definition of the term "active" or "activity" found in the specification. However, the claims at issue do not use the terms "active" or "activity." Therefore, the PTO is impermissibly importing a limitation into the claims from the specification.

Second, even if the PTO were correct to suggest that the claimed polypeptides of claims 14-17 were required to be "active," nothing in the quoted portion of the specification suggests that the "specification intends immunologically active peptides to also retain biological activity of a native or naturally-occurring PRO" as the PTO suggests. The PTO quotes the specification as stating "'biological' activity refers to a biological function (either inhibitory or stimulatory) caused by a native or naturally-occurring PRO **other than the ability to induce the production of an antibody** against an antigenic epitope possessed by a native or naturally-occurring PRO and an 'immunological' activity refers to the ability to induce the production of an antibody against an antigenic epitope possessed by a native or naturally-occurring PRO." Thus, Applicants clearly contemplated that "biological" activity was distinct from "immunological" activity. In addition, according to the PTO, the specification teaches that "'Active' or 'activity' for the purposes herein refers to form(s) of a PRO polypeptide which retain a biological **and/or** an immunological activity of native or naturally-occurring PRO." Clearly, Applicants contemplated that an "active" polypeptide can have "biological activity" **or** "immunological activity." Thus, the specification clearly teaches that a PRO polypeptide can retain "biological"

activity, which does not include immunological activity, "immunological" activity, which does not include biological activity, **or** both.

Therefore, even if Applicants have failed to disclose the "biological" activity of the PRO polypeptide as the PTO asserts, this is not relevant to the written description of the claims at issue because: (1) the claims do not recite the defined terms "active," "activity," "biological activity" or "immunological activity;" and (2) nothing in the specification requires immunologically active polypeptides to also be "biologically active."

The PTO also argues that making claimed variants is not as predictable as making nucleic acids that encode a particular amino acid sequence because "the claimed variant polypeptides are all different polypeptides ... that vary anywhere and everywhere from SEQ ID NO:46, within the metes and bounds of the recited percent identity." *Office Action* at 30. The PTO also argues that unlike biological activity, the function of being used to generate an antibody to specifically detect the polypeptide of SEQ ID NO:46 does not limit the claimed variants in any discernable, predictable or disclosed manner. *Office Action* at 30.

These arguments do not address the teaching of the *In re Wallach* case and Example 14 of the Written Description Guidelines. The *Wallach* case states that "we see no reason to require a patent applicant to list every possible permutation of the nucleic acid sequences that can encode a particular protein for which the amino acid sequence is disclosed, given the fact that it is, as explained above, a routine matter to convert back and forth between an amino acid sequence and the sequences of the nucleic acid molecules that can encode it. *In re Wallach*, 378 F.3d 1330, 1333-34 (Fed. Cir. 2004) (emphasis added). Likewise, it is a routine matter to generate the list of polypeptides which have either 95% or 99% amino acid with SEQ ID NO:46 as disclosed in the specification. Example 14 discloses that there is sufficient written description where a percent sequence identity is recited to a disclosed sequence, and a test is disclosed to determine if the variant polypeptide posses the function of the disclosed sequence. There in nothing in Example 14 that requires that the recited function limit the structure of the variant protein in any "discernable, predictable or disclosed manner." Here, Applicants have recited the function "wherein said isolated polypeptide or a fragment thereof can be used to generate an antibody which can be used to specifically detect the polypeptide of SEQ ID NO:46 in esophageal or

kidney tissue samples." Based on the disclosure of the application as filed and the skill in the art, a skilled artisan can test variant polypeptides to determine if they retain this function.

The PTO argues that "the state of the art is such that one does not typically make a variant of polypeptide in order to make antibodies that bind the polypeptide without substantially binding the variant. ... Skilled artisans would not recognize the disclosure of SEQ ID NO:46 as putting applicants in possession of the claimed genus." *Office Action* at 31.

Applicants maintain that Claims 14-17 do not require that the variant polypeptides of Claims 14-17 are capable of generating antibodies which bind the polypeptide of SEQ ID NO:46 without binding to the variant polypeptides themselves. Rather, the subject matter within the scope of Claims 14-17 includes variant polypeptides which can be used to generate antibodies which bind to both the variant polypeptide used to generate them and to the polypeptide of SEQ ID NO:46. Indeed, the PTO will appreciate that, in view of the high degree of homology between the polypeptides of Claims 14-17 and the polypeptide of SEQ ID NO:46, there are many antibodies which will bind to both polypeptides and, accordingly, the polypeptides of Claims 14-17 are useful for producing antibodies which can be used as diagnostic agents for detecting the polypeptide of SEQ ID NO:46 in a sample.

Apparently in response to Applicants citation of *Sutcliffe et al.*, Science (1983) 219:660-666 at 661-662 (previously submitted in its entirety as Exhibit 23), the PTO states:

> Applicants argue the Sutcliff abstract. It is noted that Sutcliff et al has no bearing on variants of polypeptide, but merely discusses using peptide fragments of a specific polypeptide as immunogens for generating antibodies with predefined specificity to the same polypeptide from which the peptide fragment is derived. This application is not claiming peptide immunogens or fragments of a particular protein that are used to generate antibodies to the same but larger polypeptide. *Office Action* at 31-32.

Applicants note that pending claim 14 recites "An isolated polypeptide having at least 95% amino acid sequence identity to: ... wherein said isolated polypeptide or a fragment thereof can be used to generate an antibody which can be used to specifically detect the polypeptide of SEQ ID NO:46 in esophageal or kidney tissue samples." Thus, Applicants are in fact claiming variant polypeptides that are at least 95% identical to the recited sequences, "or fragments thereof," that can be used to generate antibodies to a larger sequence. Following the teaching of Sutcliffe, one of skill in the art would know which portions of SEQ ID NO:46 would make good

fragments for generating antibodies to SEQ ID NO:46, and changes to areas outside of that region could be made without affecting the ability of that portion identified by Sutcliffe to generate an antibody to SEQ ID NO:46. As in Example 14, some testing of the variants to determine if they retain the recited function may be necessary. However, as the Guidelines indicate, disclosure of the specific sequence and the assay for testing the variants is sufficient. There is no requirement that the function limit the structure in a "discernable, predictable or disclosed manner," so long as the assay to test for the function is disclosed.

Finally, the PTO asserts that "[t]he immunological equivalent variants are not provided by disclosure of a single polypeptide because it is well established in the art [that] the retention of specificity following one or more amino acid substitutions in a polypeptide is another factor that has been shown to be unpredictable in the art." *Office Action* at 32. The PTO cites two references by McGuinness *et al.* and a reference by Houghten *et al.* as supporting this contention. Based on these references, the PTO concludes that "[t]he specification as filed does not provide written description of a representative number of variants retaining the ability to gnereat [*sic*] antibodies that specifically detect as claimed." *Office Action* at 33.

These arguments are unpersuasive because they are contrary to Sutcliffe and the PTO's Written Description Guidelines. First, Sutcliffe teaches that one of skill in the art can follow simple rules to predict which portions of a protein will make good antigens to the entire sequence. Thus, generating variants that retain the recited function is not completely "unpredictable."

Second, even if one ignores the teachings of Sutcliffe and assumes that one of skill in the art would have absolutely no idea how to make variants of SEQ ID NO:46 that retain the recited function, Example 14 of the Written Description Guidelines teaches that the claims are adequately described. There is nothing in Example 14 to suggest that making variants to a polypeptide with a particular enzymatic activity is in anyway predictable. However, the Guidelines teach that in spite of not disclosing <u>any</u> variants that retain the claimed function, or providing any guidance as to which amino acids need to be conserved, the claims in Example 14 of the Guidelines were adequately described.

The description of the hypothetical disclosure in Example 14 from the PTO's "Synopsis of Written Description Guidelines," (available at http://www.uspto.gov/web/menu/written.pdf), reads as follows:

> **Example 14: Product by Function**
>
> **Specification:** The specification exemplifies a protein isolated from liver that catalyzes the reaction of A → B. The isolated protein was sequenced and was determined to have the sequence as set forth in SEQ ID NO: 3. The specification also contemplates but does not exemplify variants of the protein wherein the variant can have any or all of the following: substitutions, deletions, insertions and additions. The specification indicates that procedures for making proteins with substitutions, deletions, insertions and additions is routine in the art and provides an assay for detecting the catalytic activity of the protein. *Synopsis of Application of Written Description Guidelines* at 53 (emphasis added).

In the analysis and conclusion of Example 14, the PTO states that:

> The single species disclosed is representative of the genus because all members have at least 95% structural identity with the reference compound and because of the presence of an assay which applicant provided for identifying all of the at least 95% identical variants of SEQ ID NO: 3 which are capable of the specified catalytic activity. ... The disclosure meets the requirements of 35 USC §112 first paragraph as providing adequate written description for the claimed invention. *Written Description Guidelines* at 54-55 (emphasis added).

The PTO's own written description guidelines state that a single species is "representative of the genus" where (1) "all members have at least 95% structural identity with the reference compound," and (2) an assay "for identifying all of the at least 95% identical variants" that retain the recited function is provided. Applicants have disclosed a single species, and both requirements (1) and (2) have been met by the specification in light of the skill in the art. Thus, the PTO's own analysis dictates the conclusion that the pending claims are adequately described by the instant specification.

The arguments accompanying the written description rejection on pages 34-37 of the Office Action are largely repetitious of those on pages 28-33, and have been addressed by Applicants' arguments above.

In conclusion, Applicants submit that they have satisfied the written description requirement for the pending claims based on the actual reduction to practice of SEQ ID NO:46, by specifying a high level of amino acid sequence identity, by describing how to make antibodies

to the disclosed sequence, all of which result in a lack of substantial variability in the species falling within the scope of the instant claims. Applicants submit that this disclosure would allow one of skill in the art to "recognize that the applicant was in possession of the necessary common attributes or features of the elements possessed by the members of the genus." Hence, Applicants respectfully request that the PTO reconsider and withdraw the written description rejection under 35 U.S.C. §112.

Rejections under 35 U.S.C. § 112, first paragraph – Enablement

The PTO further rejects pending Claims 6 and 11-17 as failing to comply with the enablement requirement. The PTO asserts that a statement assuring the availability of material deposited in the ATCC™ is required in order to enable the claimed polypeptides. The PTO has stated that the Declaration previously submitted is insufficient to obviate this rejection because it is not signed by an attorney of record over his or her registration number, and that the substitute statement provided with the last response is missing.

Applicants provide herewith a statement containing the information requested by the Examiner.

Rejections Under 35 U.S.C. §112, Second Paragraph – Indefiniteness

Claims 6, 8 and 12-17 are rejected under 35 U.S.C. §112, second paragraph, as being indefinite. The PTO states that because the specific reference to the Figure in the claim has been removed, "the claims no longer define the metes and bounds of the signal peptide as was previously referenced by specific reference to the figure in the claims. Consequently, the skilled artisan would not be readily appraised of the length of the polypeptide and would no be able to readily ascertain if they were in possession of the claimed invention." *Office Action* at 37-38 (emphasis added).

The PTO's argument is flawed because it incorrectly requires that the term "signal peptide" be defined in the claims. This is not the standard for definiteness. Instead, the test is whether "those skilled in the art would understand what is claimed when the claim is read in light of the specification." *Orthokinetics, Inc. v. Safety Travel Chairs, Inc.*, 806 F.2d 1565, 1576, 1 USPQ2d 1081, 1088 (Fed. Cir. 1986).

The specification discloses at paragraph [0072] that "Figure 46 shows the amino acid sequence (SEQ ID NO:46) derived from the coding sequence of SEQ ID NO:45 shown in Figure 45." Therefore, one of skill in the art would recognize that Figure 46 shows the entire amino acid sequence for SEQ ID NO: 46. One of skill in the art would also recognize that Figure 46 identifies particular portions of SEQ ID NO: 46, including the signal peptide sequence. Because the "signal peptide" of SEQ ID NO:46 is taught in the specification, even one not particularly skilled in the art would understand what is claimed when the claim is read in light of the specification.

"Only when a claim remains insolubly ambiguous without a discernible meaning after all reasonable attempts at construction must a court declare it indefinite." *Metabolite Labs., Inc. v. Lab. Corp. of Am. Holdings*, 370 F.3d 1354, 1366, 71 USPQ2d 1081, 1089 (Fed. Cir. 2004) (emphasis added). Claims 14-17 are not "insolubly ambiguous without discernable meaning," since any reasonable attempt to construe the claims would lead one of skill in the art to understand what is being claimed. Applicants therefore request that the rejection of claims 6, 8, and 12-17 be withdrawn.

Rejection under 35 U.S.C. § 112, first paragraph – New Matter

Claims 6, 9, 10, and 12-17 are rejected under 35 U.S.C. §112, first paragraph, for failing to comply with the written description requirement. In response to Applicants arguments which are of record and will not be repeated herein, the PTO states that:

> Applicants' arguments have been carefully considered but are not persuasive. Applicant relies solely on the description of Figure 46 to infer an extracellular domain. This is not persuasive; the term "extracellular" explicitly indicates that the region falls outside of the cell. The provision of "extracellular" domain or region does not have support in Figure 46 as filed. While the figure teaches transmembrane domains, there is no indication that the referenced membrane is the cellular membrane as opposed to the nuclear, mitochondrial, golgi or any other intracellular membrane. As such, the positioning of the intervening sequence of residues 23-223 could not necessarily be interpreted as "extracellular" as now recited. Therefore, in view of the lack of explict [*sic*] support or implicit support for the now recited metes and bounds of the extracellular domain as discussed supra, this limitation is deemed new matter. Applicants argue that the specification contemplates fragments that have the transmembrane deleted as contemplated at paragraph [0017]. This does not provide support for extracelluar [*sic*] because the recitation of transmembrane does not point one skilled in the art

> to what particular membrane and does not include intracellular membranes. The concept of a deletion mutant of a particularly defined region (transmembrane) does not proved implicit or explict [*sic*] support for other domains. Applicants also point to [0014] that teach a variety of fragments. This is not persuasive, the passage provides for fragments as specifically disclosed herein. Figure 46 only discloses and names signal peptide residues 1-22, transmembrane domain amino acids 224-250, leucine zipper pattern amino acid residues 229-251 and N-glycosylation sites. Applicants in particular rely upon "any other specifically defined fragment" littered throughout the specification. It is the position of the office that the only specifically defined fragments of Figure 46 are those recited therein. The specification does not explicitly define the claimed residues as a fragment of interest, nor does it particularly describe the residues as "extracellular". Furthermore, the inference that the metes and bounds of any particularly disclosed fragment is at the boundaries between two different fragments flies in the face of the particularly set forth N-glycosylation sites and the specification which teaches that the metes and bounds of any extracelluar [*sic*] domain can vary (see paragraph [0197]) and therefore, the specification as filed does not explicitly or inherently point to residues 23-223 as the contemplated metes and bounds of any extracelluar domain at the time of filing. *Office Action* at 38-39 (emphasis added).

The above arguments impermissibly import a limitation into the claims that is not there. In the written description rejection, the PTO states that "while the claims are read in light of the specification, specific limitations from the specification or figures are not read into the claims." *Office Action* at 37. Yet this is exactly what the PTO is doing by arguing that there is no support for an "extracellular domain." As amended, the claims recited "the amino acid sequence of amino acids 23-223 of SEQ ID NO:46," and nowhere recite the limitation "extracellular domain." Therefore, the above arguments are irrelevant, as lack of support for an "extracellular domain" which is not claimed cannot be a proper basis for rejection of the pending claims.

As Applicants have previously stated, Applicants contemplated the PRO polypeptides without their associated signal peptides and with the transmembrane domains deleted. The polypeptide that remains after the deletion of the signal peptide, (amino acids 1-22), and the transmembrane domain, (amino acids 224-250), of SEQ ID NO:46 is a polypeptide having "the amino acid sequence of amino acids 23-223 of SEQ ID NO:46," as recited in the pending claims. Whether or not this is the extracellular domain is irrelevant to the pending claims. One of skill in the art would recognize that Applicants were in possession of the claimed invention at the time of filing. Applicants therefore respectfully request that the rejection under 35 U.S.C. §112

be withdrawn.

Rejection Under 35 U.S.C. §102(b)

Pending Claims 6-7, 9, 11 and 14-15 are rejected under 35 U.S.C. § 102(b) as anticipated by STREMBL_25 database accession number Q9NY23, created October 1, 2000. The PTO asserts that the reference teaches a polypeptide sequence that has 100% sequence identity to SEQ ID NO:46. Claims 6-17 are also rejected under 35 U.S.C. § 102(b) as anticipated by Khodadoust, WO 99/67387, published December 29, 1999. The PTO asserts that Khodadoust discloses a SEQ ID NO:2 which is 100% identical to SEQ ID NO:46 of the instant application. Applicants respectfully traverse.

To be anticipated under 35 U.S.C. §102(b), the invention must be patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of the application for patent in the United States. Applicants submit that Q9NY23 and WO 99/67387 are not prior art under 35 U.S.C. §102(b) because neither was published one year before the priority date for the claimed polypeptides. The instant application is a continuation of, and claims priority under 35 U.S.C. §120 to, US Application 10/006867 filed 12/6/2001, which is a continuation of, and claims priority under 35 U.S.C. §120 to, PCT Application PCT/US00/23328 filed 8/24/2000. Applicants submit that for the reasons stated above, the claimed polypeptides have a credible, substantial, and specific utility, and are therefore entitled to a priority date of August 24, 2000.

According to the PTO, Q9NY23 was created October 1, 2000 and WO 99/67387 was published December 29, 1999. Thus, Q9NY23 was not published at least one year earlier than the August 24, 2000 priority date of the instant application, and therefore Q9NY23 and WO 99/67387 cannot be cited as prior art against the instant application under 35 U.S.C. §102(b). Applicants therefore request that the PTO reconsider and withdraw the rejection of the pending claims under 35 U.S.C. § 102(b) as anticipated by STREMBL_25, accession number Q9NY23, and Khodadoust, WO 99/67387.

Finally, Applicants note that on page 40 of the office action, the PTO makes the following incomplete argument: "Teaches the polypeptide of AAY44609 that is a human myocardium protein-7 of 335 residues in length which is 100% identical across residues 23-223

of SEQ ID NO:46. As such,." *Office Action* at 40.

Applicants request that the PTO clarify for the record whether this statement represents an additional grounds for rejection of the pending claims, and if so, which claims are rejected, under which provision of the patent statute are the claims rejected, and what the basis for rejecting the claims is. If this statement is not a further rejection of the claims, Applicants request clarification on the record that the claims are not rejected over AAY44609.

CONCLUSION

In view of the above, Applicants respectfully maintain that claims are patentable and request that they be passed to issue. Applicants invite the Examiner to call the undersigned if any remaining issues may be resolved by telephone.

Please charge any additional fees, including any fees for additional extension of time, or credit overpayment to Deposit Account No. 11-1410.

Respectfully submitted,

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Dated: Oct. 16, 2006

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